

## FBS04- Use of Alternate Light Source to Aid in Stain Identification

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### 1. Scope

- 1.1. This procedure is used to help locate possible stains of biological origin for additional testing and identification.

### 2. Background

- 2.1. An alternate light source (ALS) may be used to facilitate the visualization and detection of body fluids (e.g. saliva, semen, sweat or urine) on items of evidence. Upon illumination by the ALS at a specific wavelength, a biological stain may emit fluorescence which can be observed, marked and diagrammed. The stain can then be further characterized by using presumptive and/or confirmatory tests.

### 3. Safety

- 3.1. Wear personal protective equipment (e.g., lab coat, gloves, mask, eye protection), when carrying out standard operating procedures.
- 3.2. Read Safety Data Sheets to determine the safety hazards for chemicals and reagents used in the standard operating procedures.
- 3.3. Use alternate light source (ALS) instrumentation in a dark room.
- 3.4. Orange goggles must be worn when viewing items under ALS.

## 4. Materials Required

- 4.1. Alternate Light Source (ALS) [ROFIN Polilight PL 400 and/or Polilight-Flare Plus]
- 4.2. Orange Goggles
- 4.3. Positive Control- Semen (FBR03)
- 4.4. Negative Control- Unstained, Sterile Cotton Sheeting

## 5. Standards and Controls

- 5.1. The Positive and Negative Controls are tested prior to daily use. These results will be recorded in casework documentation.
  - 5.1.1. A known semen stain is tested as a Positive Control (FBR03). This control will exhibit a fluorescent glow when viewed under the ALS using the A450 (wavelength of 450 nm) with orange goggles.
  - 5.1.2. A sterile, unstained piece of cotton sheeting is tested as a Negative Control. This control will not exhibit a fluorescent glow when viewed under the ALS using the A450 with orange goggles.

## 6. Procedures

### Polilight PL400:

- 6.1. Turn on the main switch (fans, lamp, and power supply). Check that the fans are clear of any obstructions.
- 6.2. Check to be certain that the filter is set on the white light setting. Darken the room completely.
- 6.3. Put on orange goggles.
- 6.4. Change the filter to A450 for screening items of evidence for biological fluids (e.g. saliva, semen, sweat, or urine). Vary the angle and distance of the light source to search the item for possible stains. (**Note:** To aid in visualization, different wavelengths may be used. If using a wavelength other than 450nm, check the positive and negative controls at the alternative wavelength and document this information in the casework notes.)

- 6.5. Mark the fluorescent areas using tape, marker or some other identifier for further testing. Be aware that blood will not fluoresce, however, it may appear as a darker area when viewed under the A450.
- 6.6. When the examination is complete, change the filter back to the white light setting and turn off the main switch.
- 6.7. Note: Do not restart the lamp when the bulb is hot. Wait for the bulb to cool before restarting the power supply.

Polilight-Flare Plus:

- 6.8. Select the required head (450 nm, Blue) and insert it into the battery by aligning the red dots on both parts before pushing the two together. Push the two parts together until there is a click which indicates that the latch locating the head securely has been engaged. Lock the head in position by rotating the silver ring on the head counter clockwise by 1/8 turn.
- 6.9. Darken the room completely.
- 6.10. Put on orange goggles.
- 6.11. To switch the light ON, press the button on the battery once and release.
- 6.12. Once the light is turned on, pressing the button continuously will change the output intensity. The intensity will continue cycling in steps between 100%, to 35%, to 100%, as long as the button remains depressed.
- 6.13. To lock the flare, press the button three times in quick succession to lock the unit. After three seconds the unit can be unlocked by again pressing the button three times in quick succession.
- 6.14. Vary the angle and distance of the light source to search the item for possible stains. The strobing feature may be used to assist in visualization of stains where both the background and the evidence fluoresce. The strobing feature increases the intensity above the 100% continuous value to 150%. In some situations the contrast can be improved compared to continuous illumination. To select strobing depress the button to turn on the unit and keep the button depressed. The light will continue to strobe for as long as the button is kept depressed.

- 6.15. Mark the fluorescent areas using tape, marker or some other identifier for further testing. Be aware that blood will not fluoresce, however, it may appear as a darker area when viewed with 450 nm.
- 6.16. When the examination is complete, switch the light OFF by pressing the button on the battery once and release.
- 6.17. To charge the battery, remove the optical head from the battery unit and slide the battery in to the holder, orientating the guide on the battery so that it is aligned with the slot on the holder. When the battery is charging the ring on the charger holder glows orange. When the battery is fully charged the ring glows blue.

## **7. Sampling**

- 7.1. Not applicable

## **8. Calculations**

- 8.1. Not applicable

## **9. Uncertainty of Measurement**

- 9.1. Not applicable

## **10. Limitations**

- 10.1. It is noted that there are a variety of body fluids and substances which also fluoresce under the alternate light source. This procedure is an effective tool for locating stains to be more thoroughly tested and either included or eliminated as possible sources of DNA.

## **11. Documentation**

- 11.1. Applicable Sample Tracking and Control Software (STaCS) documentation

## **12. References**

- 12.1. Polilight PL400 Instruction Manual, ROFIN Australia Pty. Ltd.
- 12.2. Polilight Flare Plus User Guide Version 2.0c, ROFIN Australia Pty. Ltd.
- 12.3. Positive Control – Semen (FBR03)
- 12.4. Forensic Science Laboratory Quality Assurance Manual (Current Version)
- 12.5. Forensic Biology Unit Quality Assurance Manual (Current Version)
- 12.6. DFS Departmental Operations Manuals (Current Versions)
- 12.7. FSL Laboratory Operations Manuals (Current Versions)